DB Name	Query	Hit Count	Set Name
JPAB,EPAB,DWPI	dditp or (dideoxyinosine adj triphosphate)	0	L19
JPAB,EPAB,DWPI	dditp or (dideoxyinosine adj triphophate)	0	L18
JPAB,EPAB,DWPI	l15 and l16	52931	L17
JPAB,EPAB,DWPI	dna or rna or nucleic or nucleotide or oligonucleotide or polynucleotide	96396	L16
JPAB,EPAB,DWPI	sequenc$5	280046	L15
PGPB	l11 and l13	0	L14
PGPB	l11 and l1	0	L13
PGPB	dditpor (dideoxyinosine adj triphosphate)	1	L12
PGPB	l9 and l10	1375	L11
PGPB	DNA or RNA or nucleic or nucleotide or oligonucleotide or polynucleotide	1880	L10
PGPB	sequenc$5	9625	L9
USPT	l4 not l7	36	L8
USPT	l4 not l5	3	L7
USPT	l5 not l4	0	L6
USPT	l3 and l4	36	L5
USPT	dditp or (dideoxyinosine adj triphosphate)	39	L4
USPT	l1 and l2	43053	L3
USPT	Dna or rna or nucleic or nucleotide or oligonucleotide or polynucleotide	60539	L2
USPT	sequenc$5	456256	L1

WEST

Number of documents to display is limited to 50 for TI,AB,KWIC format

Generate Collection

Search Results - Record(s) 1 through 36 of 36 returned.

1. Document ID: US 6316230 B1

L8: Entry 1 of 36 File: USPT Nov 13, 2001

US-PAT-NO: 6316230
DOCUMENT-IDENTIFIER: US 6316230 B1
TITLE: Polymerase extension at 3' terminus of PNA-DNA chimera

AB: The invention provides methods and a kit for primer extension of PNA-DNA chimera from template nucleic acids using polymerases, nucleotide 5'-triphosphates, and primer extension reagents. Structural requirements of the chimera for primer extension include 5 to 15 contiguous PNA monomer units, 3 or more contiguous nucleotides, and a 3' hydroxyl terminus. The chimera and/or a nucleotide is labelled with fluorescent dyes or other labels. The methods include DNA sequencing, DNA fragment analysis, reverse transcription, mini-sequencing, chromosome labelling, amplification, and single nucleotide polymorphism (SNP) detection.

L8: Entry 1 of 36 File: USPT Nov 13, 2001

DOCUMENT-IDENTIFIER: US 6316230 B1
TITLE: Polymerase extension at 3' terminus of PNA-DNA chimera

DEPR:
Preferably the nucleotide 5'-triphosphate is ATP, dATP, ddATP, CTP, dCTP, ddCTP, GTP, dGTP, ddGTP, UTP, dUTP, TTP, dTTP, ddTTP, 5-methyl-CTP, 5-methyl-dCTP, ITP, dITP, ddITP, 2-amino-ATP, 2-amino-dATP, 7-deaza dATP, 7-deaza ddATP, 5-propynyl dCTP, 7-deaza dGTP, 7-deaza ddGTP, 5-Br-UTP, 5-Br-dUTP, 5-F-UTP, 5-F-dUTP, 5-propynyl-dUTP. Additionally, the .alpha.-phosphorus may be substituted with sulfur, as the .alpha.-thio-nucleotide 5'-triphosphates (Lee, 1992).

Full | Title | Citation | Front | Review | Classification | Date | Reference | Claims | KWIC | Draw. Desc | Image

2. Document ID: US 6306588 B1

L8: Entry 2 of 36 File: USPT Oct 23, 2001

US-PAT-NO: 6306588
DOCUMENT-IDENTIFIER: US 6306588 B1
TITLE: Polymerases for analyzing or typing polymorphic nucleic acid fragments and uses thereof

AB: The present invention provides methods for use in identifying, analyzing and typing polymorphic DNA fragments, particularly minisatellite, microsatellite or STR DNA fragments. In particular, the invention provides methods using DNA polymerases, more particularly thermostable DNA polymerases, and most particularly Thermotoga polymerases or mutants or derivatives thereof, whereby minisatellite, microsatellite or STR DNA molecules maybe amplified and analyzed for polymorphisms. The invention also relates to polymerases having reduced, substantially reduced or eliminated ability to add non-template 3' nucleotides to a synthesized nucleic acid molecule. In accordance with the invention, such reduction or elimination may be accomplished by modifying or mutating the desired polymerase.

L8: Entry 2 of 36 File: USPT Oct 23, 2001

DOCUMENT-IDENTIFIER: US 6306588 B1
TITLE: Polymerases for analyzing or typing polymorphic nucleic acid fragments and uses thereof

BSPV:
(b) incubating said mixture under conditions sufficient to synthesize nucleic acid molecules complementary to all or a portion of said templates. Such condition may include incubation with one or more deoxy- and/or dideoxyribonucleoside triphosphates. Such deoxy- and dideoxyribonucleoside triphosphates include dATP, dCTP, dGTP, dTTP, dITP, 7-deaza-dGTP, 7-deaza-dATP, dUTP, ddATP, ddCTP, ddGTP, ddITP, ddTTP, [.alpha.-S]dATP, [.alpha.-S]dTTP, [.alpha.-S]dGTP, and [.alpha.-S]dCTP. The synthesized nucleic acid molecules may in accordance with the invention be cloned into one or more vectors.

BSPV:
(d) separating said synthesized DNA molecules by size so that at least a part of the nucleotide sequence of said first DNA molecule can be determined. Such terminator nucleotides include but are not limited to dideoxyribonucleoside triphosphates such as ddTTP, ddATP, ddGTP, ddITP or ddCTP.

DEPR:
Nucleotide. As used herein "nucleotide" refers to a base-sugar-phosphate combination. Nucleotides are monomeric units of a nucleic acid sequence (DNA and RNA). The term nucleotide includes deoxyribonucleoside triphosphates such as dATP, dCTP, dITP, dUTP, dGTP, dTTP, or derivatives thereof. Such derivatives include, for example, [.alpha.S]dATP, 7-deaza-dGTP and 7-deaza-dATP. The term nucleotide as used herein also refers to dideoxyribonucleoside triphosphates (ddNTPs) and their derivatives. Illustrated examples of dideoxyribonucleoside triphosphates include, but are not limited to, ddATP, ddCTP, ddGTP, ddITP, and ddTTP. According to the present invention, a "nucleotide" may be unlabeled or detectably labeled by well known techniques. Detectable labels include, for example, radioactive isotopes, fluorescent labels, chemiluminescent labels, bioluminescent labels and enzyme labels.

Full | Title | Citation | Front | Review | Classification | Date | Reference KWIC | Draw Desc | Image

3. Document ID: US 6277608 B1

L8: Entry 3 of 36 File: USPT Aug 21, 2001

US-PAT-NO: 6277608
DOCUMENT-IDENTIFIER: US 6277608 B1
TITLE: Recombinational cloning using nucleic acids having recombination sites

AB: Recombinational cloning is provided by the use of nucleic acids, vectors and methods, in vitro and in vivo, for moving or exchanging segments of DNA molecules using engineered recombination sites and recombination proteins to provide chimeric DNA molecules that have the desired characteristic(s) and/or DNA segment(s).

L8: Entry 3 of 36 File: USPT Aug 21, 2001

DOCUMENT-IDENTIFIER: US 6277608 B1
TITLE: Recombinational cloning using nucleic acids having recombination sites

DEPR:
Nucleotide: refers to a base-sugar-phosphate combination. Nucleotides are monomeric units of a nucleic acid sequence (DNA and RNA). The term nucleotide includes ribonucleoside triphosphatase ATP, UTP, CTG, GTP and deoxyribonucleoside triphosphates such as dATP, dCTP, dITP, dUTP, dGTP, dTTP, or derivatives thereof Such derivatives include, for example, [.alpha.S]dATP, 7-deaza-dGTP and 7-deaza-dATP. The term nucleotide as used herein also refers to dideoxyribonucleosidetriphosphates (ddNTPs) and their derivatives. Illustrated examples of dideoxyribonucleoside triphosphates include, but are not limited to, ddATP, ddCTP, ddGTP, ddITP, and ddTTP. According to the present invention, a "nucleotide" may be unlabeled or detectably labeled by well known techniques. Detectable labels include, for example, radioactive isotopes, fluorescent labels, chemiluminescent labels, bioluminescent labels and enzyme labels.

Full | Title | Citation | Front | Review | Classification | Date | Reference KWIC | Draw Desc | Image

---

4. Document ID: US 6274738 B1

L8: Entry 4 of 36 File: USPT Aug 14, 2001

US-PAT-NO: 6274738
DOCUMENT-IDENTIFIER: US 6274738 B1
TITLE: Carboxamide derivatives having aryl and thiazole rings

AB: The invention provides DNA primase assays suitable for identifying DN primase modulating agents, methods of modulating DNA primase activity, compounds for modulating DNA primase activity, and compositions which modulate DNA primase.

L8: Entry 4 of 36 File: USPT Aug 14, 2001

DOCUMENT-IDENTIFIER: US 6274738 B1
TITLE: Carboxamide derivatives having aryl and thiazole rings

BSPR:
In a variation, the reaction further comprises a DNA polymerase activity, typically a mammalian DNA polymerase .alpha., and reaction conditions suitable for catalytic activity of the DNA polymerase(s), such that product polynucleotides formed by the activity of the DNA primase may be extended further by the DNA polymerase, which can be useful to enhance or amplify the signal resulting from incorporation of labeled nucleotide into product polynucleotide. In such embodiments, one or more labeled dNTPs are also present in the reaction mixture, typically including each dNTP which would be present in a complementary strand of a template polynucleotide. In an embodiment, only one species of dNTP is labeled (e.g., .sup.32 P-.alpha.-dCTP or biotinylated dGTP). In an alternative embodiment, multiple species of dNTP are labeled, and in a variation all dNTP species are labeled. In general, the method employs a DNA primase and/or DNA primase/DNA polymerase reaction comprising: (1) a template polynucleotide capable of providing a template for a mammalian DNA primase; (2) a labeled nucleotide or polynucleotide species, and optionally (for heteronucleotide template sequences)

unlabeled nucleotide species such that the reaction contains nucleotide species (either labeled and unlabeled) representing nucleotides which can be efficiently incorporated in a complementary strand to the template polynucleotide; (3) a predetermined amount of mammalian, preferably human, DNA primase and, optionally, a predetermined amount of a mammalian, preferably human, DNA polymerase (e.g., DNA pol .alpha.) in suitable reaction conditions (e.g., pH, ionic strength, ATP, temperature, metal ion concentration, etc.). The DNA primase reaction typically contains either: (1) at least one labeled ribonucleotide species, and optionally additional unlabeled ribonucleotide species necessary for synthesis of a complementary RNA strand to the template, for direct reporting of RNA synthesis as a measure of primase activity, or (2) at least one labeled deoxyribonucleotide species, and optionally additional unlabeled ribonucleotide species and deoxyribonucleotide species necessary for synthesis of a complementary strand to the template, for reporting of DNA synthesis which indirectly reports primase activity as RNA primer synthesis is necessary for initiation of DNA synthesis. In embodiments where labeled DNA synthesis is reported, reaction times can be shortened to reduce the average length of DNA chains synthesized and/or a DNA chain terminator nucleotide (such as, e.g., dideoxynucleotide: ddCTP, ddTTP, ddGTP, ddATP, ddITP, etc.) can be included in sufficient amount to reduce average DNA chain length which may be preferable to ensure that DNA synthesis is substantially proportional to RNA primer synthesis (and that the readout, whether by labeled RNA synthesis or labeled DNA synthesis, is representative or proportional to DNA primase activity in the reaction).

BSPR:
In a variation, labeled nucleotides bear distinct labels to distinguish template versus non-template directed polymerization in a DNA primase reaction or coupled DNA primase/DNA polymerase reaction. A first labeled nucleotide species having a first label is incorporated in polynucleotides produced from template-directed polynucleotide synthesis, such as DNA primase-catalyzed oligoribonucleotide primer synthesis or DNA primase/DNA polymerase-catalyzed elongation of a oligoribonucleotide primer by template-directed polymerization. A second labeled nucleotide species having a second label which can be distinguished or discriminated (i.e., is separately detectable) from the first label of the first nucleotide species is incorporated substantially only in polynucleotides produced by untemplated polymerization. In this variation, a "nucleotide deficient template" serves as a primase template, and is a homopolymer or a heteropolymer polynucleotide composed of residues of two or three deoxyribonucleotide species (i.e., the template lacks at least one DNTP species) wherein at least one of said deoxyribonucleotide residues is a complement nucleotide of the first labeled nucleotide, and wherein none of said deoxyribonucleotide residues is a complement nucleotide of said second labeled nucleotide, whereby template-directed polynucleotide synthesis by DNA primase or DNA primase/DNA polymerase yields a product polynucleotide comprising an incorporated (i.e., polymerized) residue of said first labeled nucleotide species and substantially lacking incorporated residues of said second labeled nucleotide species (except for minor misincorporation errors inherent in polynucleotide polymerases). The second labeled polynucleotide species is complementary to a dNTP species which is not present in the nucleotide-deficient template, and therefore polynucleotide products of the reaction having incorporated second labeled nucleotide residues substantially represent reaction products generated by untemplated polymerization. The method employs a DNA primase and/or DNA primase/DNA polymerase reaction comprising: (1) a nucleotide-deficient template and substantially lacking other template species; (2) a first labeled nucleotide species and a second labeled nucleotide species, and optionally unlabeled nucleotide species such that the reaction contains nucleotide species (either labeled and unlabeled) representing nucleotides which can be efficiently incorporated in a complementary strand to the nucleotide-deficient template; (3) a predetermined amount of mammalian, preferably human, DNA primase and, optionally, a predetermined amount of a mammalian, preferably human, DNA polymerase (e.g., DNA pol a) in suitable reaction conditions (e.g., pH, ionic strength, ATP, temperature, metal ion concentration, etc.). The DNA primase reaction typically contains a first and second labeled nucleotide species which are either both ribonucleotides or are both deoxyribonucleotides. In embodiments where labeled DNA synthesis is reported, reaction times can be shortened, as described supra, to reduce the average length of DNA chains synthesized and/or a DNA chain terminator nucleotide (dideoxynucleotide: ddCTP, ddTTP, ddGTP, ddATP, ddITP, etc.) is included in sufficient amount to reduce average DNA chain length such that DNA synthesis is substantially proportional to RNA primer synthesis

(i.e., the readout, whether labeled RNA synthesis or labeled DNA synthesis, is representative or proportional to DNA primase activity in the reaction). Alternatively, or in combination, the product polynucleotide(s) can be detected by hybridization with a complementary strand probe polynucelotide which may be labeled and/or which may be immobilized and used to capture, by hybridization, a labeled product polynucelotide having sufficient complementarity to hybridize under suitable hybridization conditions.

Full | Title | Citation | Front | Review | Classification | Date | Reference KWIC | Draw Desc | Image

---

5. Document ID: US 6274353 B1

L8: Entry 5 of 36 File: USPT Aug 14, 2001

US-PAT-NO: 6274353
DOCUMENT-IDENTIFIER: US 6274353 B1
TITLE: Method and compositions for improved polynucleotide synthesis

AB: The sensitivity and specificity of polynucleotide synthesis is increased by protecting the 3'-end of an oligonucleotide used as a primer in the synthesis of the polynucleotide. Protection of the 3'-end of an oligonucleotide prevents non-specific chain elongation. Removal of blocking group an elevated temperature, using a thermostable enzyme, permits template-specific polynucleotide synthesis. The present invention also provides oligonucleotides with a 3' end protected by a blocking group and a thermostable enzyme capable of removing the blocking group at an elevated temperature. The compositions and methods of the invention are very useful in a variety of techniques for DNA/RNA amplification and analysis, including medical genetics research and diagnosis, pathogen detection, forensic, and animal and plant genetics applications, among others.

L8: Entry 5 of 36 File: USPT Aug 14, 2001

DOCUMENT-IDENTIFIER: US 6274353 B1
TITLE: Method and compositions for improved polynucleotide synthesis

DEPR:
Nucleotide: As used herein "nucleotide" refers to a base-sugar-phosphate combination. Nucleotides are monomeric units of a nucleic acid sequence (DNA and RNA). The term nucleotide includes deoxyribonucleoside triphosphates such as dATP, dITP, dUTP, dGTP, dTTP, or derivatives thereof. Such derivatives include for example, (.alpha.S) dATP, 7-deaza-dGTP and 7-deaza-dATP. The term nucleotide as used herein also include dideoxyribonucleoside triphosphates (ddNTPs) and their derivatives. Illustrated examples of dideoxyribonucleoside triphosphates include, but are not limited to, ddATP, ddCTP, ddGTP, ddITP, and ddTTP. According to the present invention, a "nucleotide" may be unlabeled or detectably labeled by well known techniques. Detectable labels include, for example, radioactive isotopes, fluorescent labels, chemiluminescent labels and enzyme labels.

Full | Title | Citation | Front | Review | Classification | Date | Reference KWIC | Draw Desc | Image

---

6. Document ID: US 6140086 A

L8: Entry 6 of 36 File: USPT Oct 31, 2000

US-PAT-NO: 6140086
DOCUMENT-IDENTIFIER: US 6140086 A
TITLE: Methods and compositions for cloning nucleic acid molecules

AB: The present invention is directed generally to methods facilitating the cloning of nucleic acid molecules. In particular, the invention relates to the use of polymerase inhibitors, including but not limited to anti-polymerase antibodies (such as anti-Taq antibodies) and fragments thereof, to inactivate residual polymerase activity remaining after the amplification (particularly via PCR) of a target nucleic acid molecule. The invention further provides compositions, particularly storage-stable compositions, comprising one or more components, such as one or more restriction endonucleases and one or more polymerase inhibitors, that are useful in cloning amplified or synthesized nucleic acid molecules by the above-described methods. The invention also relates to nucleic acid molecules produced by these methods, and to genetic constructs (such as vectors) and host cells comprising these nucleic acid molecules.

L8: Entry 6 of 36 File: USPT Oct 31, 2000

DOCUMENT-IDENTIFIER: US 6140086 A
TITLE: Methods and compositions for cloning nucleic acid molecules

DEPR:
As used herein, "nucleotide" refers to a base-sugar-phosphate combination. Nucleotides are monomeric units of a nucleic acid sequence (DNA and RNA). The term nucleotide includes deoxyribonucleoside triphosphates ("dNTPs") such as dATP, dCTP, dITP, dUTP, dGTP, dTTP, or derivatives thereof. Such derivatives include, for example, [.alpha.S]dATP, 7-deaza-dGTP and 7-deaza-dATP. The term nucleotide as used herein also refers to dideoxyribonucleoside triphosphates ("ddNTPs") and their derivatives, including, but not limited to, ddATP, ddCTP, ddGTP, ddITP, and ddTTP. In addition, the term nucleotide includes ribonucleoside triphosphates (rNTPs) such as rATP, rCTP, rITP, rUTP, rGTP, rTTP and their derivatives, which are analogous to the above-described dNTPs and ddNTPs except that the rNTPs comprise ribose instead of deoxyribose or dideoxyribose in their sugar-phosphate backbone. According to the present invention, a "nucleotide" may be unlabeled or detectably labeled by well known techniques. Detectable labels include, for example, radioactive isotopes, fluorescent labels, chemilurninescent labels, bioluminescent labels and enzyme labels.

Full | Title | Citation | Front | Review | Classification | Date | Reference KWIC | Draw Desc | Image

7. Document ID: US 6096499 A

L8: Entry 7 of 36 File: USPT Aug 1, 2000

US-PAT-NO: 6096499
DOCUMENT-IDENTIFIER: US 6096499 A
TITLE: Mammalian DNA primase screen and activity modulating agents

AB: The invention provides DNA primase assays suitable for identifying DNA primase modulating agents, methods of modulating DNA primase activity and compositions which modulate DNA primase.

L8: Entry 7 of 36 File: USPT Aug 1, 2000

DOCUMENT-IDENTIFIER: US 6096499 A
TITLE: Mammalian DNA primase screen and activity modulating agents

BSPR:
In a variation, the reaction further comprises a DNA polymerase activity, typically a mammalian DNA polymerase .alpha., and reaction conditions suitable for catalytic activity of the DNA polymerase(s), such that product polynucleotides formed by the activity of the DNA primase may be extended further by the DNA polymerase, which can be useful to enhance or amplify the signal resulting from incorporation of labeled nucleotide into product polynucleotide. In such embodiments, one or more labeled dNTPs are also present in the reaction mixture, typically including each dNTP which would be present in a complementary strand of a template polynucleotide. In an embodiment, only one species of dNTP is labeled (e.g., .sup.32 P-.alpha.-dCTP or biotinylated dGTP). In an alternative embodiment, multiple species of dNTP are labeled, and in a variation all dNTP species are labeled. In general, the method employs a DNA primase and/or DNA primase/DNA polymerase reaction comprising: (1) a template polynucleotide capable of providing a template for a mammalian DNA primase; (2) a labeled nucleotide or polynucleotide species, and optionally (for heteronucleotide template sequences) unlabeled nucleotide species such that the reaction contains nucleotide species (either labeled and unlabeled) representing nucleotides which can be efficiently incorporated in a complementary strand to the template polynucleotide; (3) a predetermined amount of mammalian, preferably human, DNA primase and, optionally, a predetermined amount of a mammalian, preferably human, DNA polymerase (e.g., DNA pol .alpha.) in suitable reaction conditions (e.g., pH, ionic strength, ATP, temperature, metal ion concentration, etc.). The DNA primase reaction typically contains either: (1) at least one labeled ribonucleotide species, and optionally additional unlabeled ribonucleotide species necessary for synthesis of a complementary RNA strand to the template, for direct reporting of RNA synthesis as a measure of primase activity, or (2) at least one labeled deoxyribonucleotide species, and optionally additional unlabeled ribonucleotide species and deoxyribonucleotide species necessary for synthesis of a complementary strand to the template, for reporting of DNA synthesis which indirectly reports primase activity as RNA primer synthesis is necessary for initiation of DNA synthesis. In embodiments where labeled DNA synthesis is reported, reaction times can be shortened to reduce the average length of DNA chains synthesized and/or a DNA chain terminator nucleotide (such as, e.g., dideoxynucleotide: ddCTP, ddTTP, ddGTP, ddATP, ddITP, etc.) can be included in sufficient amount to reduce average DNA chain length which may be preferable to ensure that DNA synthesis is substantially proportional to RNA primer synthesis (and that the readout, whether by labeled RNA synthesis or labeled DNA synthesis, is representative or proportional to DNA primase activity in the reaction).

BSPR:
representing nucleotides which can be efficiently incorporated in a complementary strand to the nucleotide-deficient template; (3) a predetermined amount of mammalian, preferably human, DNA primase and, optionally, a predetermined amount of a mammalian, preferably human, DNA polymerase (e.g., DNA pol .alpha.) in suitable reaction conditions (e.g., pH, ionic strength, ATP, temperature, metal ion concentration, etc.). The DNA primase reaction typically contains a first and second labeled nucleotide species which are either both ribonucleotides or are both deoxyribonucleotides. In embodiments where labeled DNA synthesis is reported, reaction times can be shortened, as described supra, to reduce the average length of DNA chains synthesized and/or a DNA chain terminator nucleotide (dideoxynucleotide: ddCTP, ddTTP, ddGTP, ddATP, ddITP, etc.) is included in sufficient amount to reduce average DNA chain length such that DNA synthesis is substantially proportional to RNA primer synthesis (i.e., the readout, whether labeled RNA synthesis or labeled DNA synthesis, is representative or proportional to DNA primase activity in the reaction). Alternatively, or in combination, the product polynucleotide(s) can be detected by hybridization with a complementary strand probe polynucelotide which may be labeled and/or which may be immobilized and used to capture, by hybridization, a labeled product polynucelotide having sufficient complementarity to hybridize under suitable hybridization conditions.

8. Document ID: US 6043036 A

L8: Entry 8 of 36 File: USPT Mar 28, 2000

US-PAT-NO: 6043036
DOCUMENT-IDENTIFIER: US 6043036 A
TITLE: Method of sequencing nucleic acids by shift registering

AB: The present invention describes a method of sequencing nucleic acids in which mixtures of oligonucleotide fragments are derived from sequencing reactions using combinations of the 2',3'-dideoxynucleoside 5'-triphosphate or 3' deoxynucleoside 5'-triphosphate terminators and appropriate concentrations of four dNTPs (2'-deoxynucleoside 5' triphosphates, e.g., dATP, dCTP, dGTP, dTTP, dITP, 7-deaza-GTP). These fragments are generated by enzymatic extension of a primer hybridized to the single-stranded template DNA to be sequenced. In contrast to common slab gel sequencing methods, the method of the instant invention does not require precise alignment of the four separation sets of the terminated fragments to permit deduction of the DNA sequence. In addition, the method possesses inherent redundancy in the separations, which facilitates sequence assignment by resolving sequence uncertainties or anomalies.

L8: Entry 8 of 36 File: USPT Mar 28, 2000

DOCUMENT-IDENTIFIER: US 6043036 A
TITLE: Method of sequencing nucleic acids by shift registering

DEPR:
Another useful implementation of the instant invention is based on six sets of labeled oligonucleotide fragments produced from the template DNA in a set of six reaction mixtures. Set one is comprised of labeled oligonucleotide fragments produced with ddA and ddC terminators, set two is comprised of labeled oligonucleotide fragments produced with ddA and ddG terminators, set three is comprised of labeled oligonucleotide fragments produced from ddA and ddT terminators, set four is comprised of labeled oligonucleotide fragments produced from ddC and ddG terminators, set five is comprised of labeled oligonucleotide fragments produced from ddC and ddT terminators, and set six is comprised of labeled oligonucleotide fragments produced from ddG and ddT terminators. These labeled fragments are separated according to size, each set determined independently by performing the separations in a temporally or spatially distinct manner. Detection of the distinct pattern derived from each set of separations produces a pattern of the linear order of one of the nucleotides from the template DNA, and these patterns can be compared and combined with each other to deduce the template sequence. Yet another useful implementation is based on a set of four reaction mixtures selected from the following six possible combinations: combination one being comprised of labeled oligonucleotide fragments produced with ddA and ddC terminators, combination two being comprised of labeled oligonucleotide fragments produced with ddA and ddG terminators, combination three being comprised of labeled oligonucleotide fragments produced from ddA and ddT terminators, combination four being comprised of labeled oligonucleotide fragments produced from ddC and ddG terminators, combination five being comprised of labeled oligonucleotide fragments produced from ddC and ddT terminators, and combination six being comprised of labeled oligonucleotide fragments produced from ddG and ddT terminators. Each of the terminators ddA, ddC, ddG, and ddT is represented in two of the four reaction mixtures. For example, one possible combination of four reaction mixtures would be ddA/ddC, ddC/ddG, ddG/ddT, and ddT/ddA. Yet another possible combination of four mixtures would be ddA/ddG, ddC/ddT, ddA/ddC, and ddG/ddT. There are at least four useful combinations using this strategy. In all cases recited above, alternative 2',3'-dideoxynucleoside 5'-triphosphate terminators, such as 2',3'-dideoxyinosine 5'-triphosphate (ddITP), modified analogs of 2',3'-dideoxynucleoside 5'-triphosphate terminators such as fluorophore-modified (DyeDeoxy.TM. Terminators, Perkin-Elmer/ABI) or biotin-modified (GATC, Konstanz, Germany) terminators or 3'-deoxynucleoside 5'-triphosphate terminators or analogs thereof, could also be utilized.

9. Document ID: US 6015668 A

L8: Entry 9 of 36 File: USPT Jan 18, 2000

US-PAT-NO: 6015668
DOCUMENT-IDENTIFIER: US 6015668 A
TITLE: Cloned DNA polymerases from thermotoga and mutants thereof

AB: The invention relates to a substantially pure thermostable DNA polymerase from Thermotoga (Tne and Tma) and mutants thereof. The Tne DNA polymerase has a molecular weight of about 100 kilodaltons and is more thermostable than Taq DNA polymerase. The mutant DNA polymerase has at least one mutation selected from the group consisting of (1) a first mutation that substantially reduces or eliminates 3'.fwdarw.5' exonuclease activity of said DNA polymerase; (2) a second mutation that substantially reduces or eliminates 5'.fwdarw.3' exonuclease activity of said DNA polymerase; (3) a third mutation in the O helix of said DNA polymerase resulting in said DNA polymerase becoming non-discriminating against dideoxynucleotides. The present invention also relates to the cloning and expression of the wild type or mutant DNA polymerases in E. coli, to DNA molecules containing the cloned gene, and to host cells which express said genes. The DNA polymerases of the invention may be used in well-known DNA sequencing and amplification reactions.

L8: Entry 9 of 36 File: USPT Jan 18, 2000

DOCUMENT-IDENTIFIER: US 6015668 A
TITLE: Cloned DNA polymerases from thermotoga and mutants thereof

BSPV:
(d) separating said synthesized DNA molecules by size so that at least a part of the nucleotide sequence of said first DNA molecule can be determined. Such terminator nucleotides include ddTTP, ddATP, ddGTP, ddITP or ddCTP.

DEPR:
Nucleotide. As used herein "nucleotide" refers to a base-sugar-phosphate combination. Nucleotides are monomeric units of a nucleic acid sequence (DNA and RNA). The term nucleotide includes deoxyribonucleoside triphosphates such as dATP, dCTP, dITP, dUTP, dGTP, dTTP, or derivatives thereof. Such derivatives include, for example, [.alpha.S]dATP, 7-deaza-dGTP and 7-deaza-dATP. The term nucleotide as used herein also refers to dideoxyribonucleoside triphosphates (ddNTPs) and their derivatives. Illustrated examples of dideoxyribonucleoside triphosphates include, but are not limited to, ddATP, ddCTP, ddGTP, ddITP, and ddTTP. According to the present invention, a "nucleotide" may be unlabeled or detectably labeled by well known techniques. Detectable labels include, for example, radioactive isotopes, fluorescent labels, chemiluminescent labels, bioluminescent labels and enzyme labels.

DEPR:
The wild type and mutant Thermotoga DNA polymerases (e.g., Tma and Tne) of the present invention may be used in well known DNA sequencing, DNA labeling, DNA amplification and CDNA synthesis reactions. Thermotoga DNA polymerase mutants devoid of or substantially reduced in 3'.fwdarw.5' exonuclease activity, devoid of or substantially reduced in 5'.fwdarw.3' exonuclease activity, or containing one or mutations in the O-helix that make the enzyme nondiscriminatory for dNTPs and ddNTPs (e.g., a Phe.sup.730 .fwdarw.Tyr.sup.730 mutation of SEQ ID NO: 3) are especially useful for DNA sequencing, DNA labeling, and DNA amplification reactions and CDNA synthesis. Moreover, Thermotoga DNA polymerase mutants containing two or more of these properties are also especially useful for DNA sequencing, DNA labeling, DNA amplification or cDNA synthesis reactions. As is well known, sequencing reactions (isothermal DNA sequencing and cycle sequencing

well known, sequencing reactions (isothermal DNA sequencing and cycle sequencing of DNA) require the use of DNA polymerases. Dideoxy-mediated sequencing involves the use of a chain-termination technique which uses a specific polymer for extension by DNA polymerase, a base-specific chain terminator and the use of polyacrylamide gels to separate the newly synthesized chain-terminated DNA molecules by size so that at least a part of the nucleotide sequence of the original DNA molecule can be determined. Specifically, a DNA molecule is sequenced by using four separate DNA sequence reactions, each of which contains different base-specific terminators. For example, the first reaction will contain a G-specific terminator, the second reaction will contain a T-specific terminator, the third reaction will contain an A-specific terminator, and a fourth reaction may contain a C-specific terminator. Preferred terminator nucleotides include dideoxyribonucleoside triphosphates (ddNTPs) such as ddATP, ddTTP, ddGTP, ddITP and ddCTP. Analogs of dideoxyribonucleoside triphosphates may also be used and are well known in the art.

CLPR:
3. The method of sequencing as claimed in claim 1, wherein said terminator nucleotide is selected from the group consisting of: ddTTP, ddATP, ddGTP, ddITP, and ddCTP.

CLPR:
48. The method of sequencing as claimed in claim 46, wherein said terminator nucleotide is selected from the group consisting of: ddTTP, ddATP, ddGTP, ddITP, or ddCTP.

☐ 10. Document ID: US 6007989 A

L8: Entry 10 of 36 File: USPT Dec 28, 1999

US-PAT-NO: 6007989
DOCUMENT-IDENTIFIER: US 6007989 A
TITLE: Methods of screening for compounds that derepress or increase telomerase activity

AB: Method and compositions are provided for the determination of telomere length and telomerase activity, as well as the ability to increase or decrease telomerase activity in the treatment of proliferative diseases. Particularly, primers are elongated under conditions which minimize interference from other genomic sequences, so as to obtain accurate determinations of telomeric length or telomerase activity. In addition, compositions are provided for intracellular inhibition of telomerase activity and means are shown for slowing or reversing the loss of telomeric repeats in aging cells.

L8: Entry 10 of 36 File: USPT Dec 28, 1999

DOCUMENT-IDENTIFIER: US 6007989 A
TITLE: Methods of screening for compounds that derepress or increase telomerase activity

DRPR:
FIGS. 20A and B show the effect of nucleoside triphosphate analogs on pausing patterns and processivity of telomerase in vitro. Specifically, FIG. 20A shows telomerase reactions in the presence or absence of the indicated nucleoside triphosphate analogs. Unlabeled TTP competitor was also analyzed as a control, with and without primer in the reaction mix. Products were then analyzed on a denaturing polyacrylamide gel. FIG. 20B shows standard telomerase reactions were performed in the presence of ddGTP (lanes 4-6), ddITP (lanes 7-9), or DMSO (lane 1). DMSO was the solvent for ddGTP and at the highest concentration tested (1%) showed no effect on the reactions compared with control reactions run without analog or DMSO (control lanes 2-3). Products were analyzed on a denaturing

polyacrylamide gel.

DEPR:
We analyzed the effects of various nucleoside analogs, which are chain-terminating inhibitors of retroviral reverse transcriptases, on Tetrahymena thermophila telomerase activity in vitro, and on telomere length and maintenance, cell division and conjugation of Tetrahymena cells in vivo. In vitro assays of telomerase activity showed that arabinofuranyl-guanosine triphosphate (Ara-GTP) and ddGTP were both very efficient inhibitors of incorporation of labeled nucleotides into telomeric DNA repeats, even at low inhibitor concentrations, while azidothymidine triphosphate (AZT-TP), dideoxyinosine triphosphate (ddITP) or ddTTP were less efficient inhibitors of incorporation. All of these nucleoside triphosphate analogs, however, produced analog-specific alterations of the normal banding patterns seen upon gel electrophoresis of the synthesis products of telomerase, suggesting that the competitive and/or chain terminating action differed at different positions along the RNA template.

DEPR:
We also tested the effects of dideoxynucleoside triphosphates (ddNTPs) on the telomerase reaction. As shown previously for telomerase [Greider 43 Cell, 405, 1985], and as is the case for many other reverse trancriptases, ddNTPs are recognized by the enzyme and incorporated, causing chain termination with a subsequent shift in banding patterns and reduction of the average product length. Consistent with previous qualitative analyses of Tetrahymena and human telomerases [Greider 43 Cell, 405, 1985; Morin 59 Cell, 521, 1989], ddGTP and ddTTP each inhibited the incorporation of labeled .sup.32 P-NTP into elongation products (FIG. 19D and E). ddGTP was a much more efficient inhibitor than ddTTP: under these reaction conditions 50% inhibition occurred at <0.1 and 5 .mu.M ddGTP and ddTTP respectively. As observed previously for Tetrahymena telomerase [Greider 43 Cell, 405, 1985], no significant effects were seen with either ddCTP or ddATP. In addition, ddITP inhibited telomerase (FIG. 19E), although less efficiently than ddGTP, with 50% inhibition occurring at 3 .mu.M ddITP.

DEPR:
The size distribution of labeled products was then analyzed by denaturing polyacrylamide gel electrophoresis. Consistent with the expectation for a chain-terminator, the proportion of longer telomerase products was decreased in the presence of AZT compared with cold TTP competitor controls (FIG. 20A; compare lanes 1 and 2 with lanes 3 to 5), and in the presence of Ara-G (FIG. 20A; lanes 7 and 8). Average product length also decreased in the presence of Ara-GTP, ddGTP and ddITP (FIG. 20A and B). In addition, each nucleoside triphosphate analog produced distinctive and characteristic patterns of chain termination, as shown by analysis of the shifts in the banding patterns of the elongation products. With AZT-triphosphate, we saw increased relative intensities of the bands corresponding to the incorporation of T residues (copying the A residues at positions 2 and 3 on the template RNA (see FIG. 18A)). This change in banding pattern is consistent with simple chain termination, which is predicted to increase the intensity of bands corresponding to the position of both incorporated T residues. Similar effects were seen with ddTTP. We interpret this to mean that AZT-triphosphate was recognized by the enzyme and incorporated into the correct positions in the growing telomeric sequence, causing chain termination. However it cannot be excluded that the increase in relative intensity of the band corresponding to position 3 on the template, which precedes addition of the second T, is also attributable to pausing caused by competition with TTP and a slower reaction rate with AZT-triphosphate at position 2. Should AZT-triphosphate, or related nucleotide analogs, be incorporated into telomeric DNA where they would not be incorporated by DNA polymerase into other DNA, then such nucleotide analogs may be used to kill telomerase positive cells by causing them to generate telomeric DNA toxic to the cell, or at least altered in such a way that telomerase-mediated cell immortalization was inhibited.

DEPR:
The results with Ara-GTP were also consistent with incorporation of Ara-G and consequent chain termination (FIG. 20A, lanes 7 and 8). Although there are four positions at which a G residue can be incorporated and therefore at which chain termination could occur, the strongest increase was in the band corresponding to the G residue specified by position 4, in the middle of the telomerase RNA template (see FIG. 18A). With ddGTP, chain termination appeared to occur most efficiently at positions 6 and 5 (FIG. 20B, compare lane 1 with lanes 4 to 6),

and with ddITP, at position 5 (lanes 7 to 9).

---

11. Document ID: US 5948614 A

L8: Entry 11 of 36 File: USPT Sep 7, 1999

US-PAT-NO: 5948614
DOCUMENT-IDENTIFIER: US 5948614 A
TITLE: Cloned DNA polymerases from thermotoga maritima and mutants thereof

AB: The invention relates to a substantially pure thermostable DNA polymerase from Thermotoga (Tne and Tma) and mutants thereof. The Tne DNA polymerase has a molecular weight of about 100 kilodaltons and is more thermostable than Taq DNA polymerase. The mutant DNA polymerase has at least one mutation selected from the group consisting of (1) a first mutation that substantially reduces or eliminates 3'.fwdarw.5' exonuclease activity of said DNA polymerase; (2) a second mutation that substantially reduces or eliminates 5'.increment.3' exonuclease activity of said DNA polymerase; (3) a third mutation in the O helix of said DNA polymerase resulting in said DNA polymerase becoming non-discriminating against dideoxynucleotides. The present invention also relates to the cloning and expression of the wild type or mutant DNA polymerases in E. coli, to DNA molecules containing the cloned gene, and to host cells which express said genes. The DNA polymerases of the invention may be used in well-known DNA sequencing and amplification reactions.

L8: Entry 11 of 36 File: USPT Sep 7, 1999

DOCUMENT-IDENTIFIER: US 5948614 A
TITLE: Cloned DNA polymerases from thermotoga maritima and mutants thereof

BSPV:
(d) separating said synthesized DNA molecules by size so that at least a part of the nucleotide sequence of said first DNA molecule can be determined. Such terminator nucleotides include ddTTP, ddATP, ddGTP, ddITP or ddCTP.

DEPR:
Nucleotide. As used herein "nucleotide" refers to a base-sugar-phosphate combination. Nucleotides are monomeric units of a nucleic acid sequence (DNA and RNA). The term nucleotide includes deoxyribonucleoside triphosphates such as dATP, dCTP, dITP, dUTP, dGTP, dTTP, or derivatives thereof. Such derivatives include, for example, [.alpha.S]dATP, 7-deaza-dGTP and 7-deaza-dATP. The term nucleotide as used herein also refers to dideoxyribonucleoside triphosphates (ddNTPs) and their derivatives. Illustrated examples of dideoxyribonucleoside triphosphates include, but are not limited to, ddATP, ddCTP, ddGTP, ddITP, and ddTTP. According to the present invention, a "nucleotide" may be unlabeled or detectably labeled by well known techniques. Detectable labels include, for example, radioactive isotopes, fluorescent labels, chemiluminescent labels, bioluminescent labels and enzyme labels.

DEPR:
The wild type and mutant Thermotoga DNA polymerases (e.g., Tma and Tne) of the present invention may be used in well known DNA sequencing, DNA labeling, DNA amplification and cDNA synthesis reactions. Thermotoga DNA polymerase mutants devoid of or substantially reduced in 3'.fwdarw.5' exonuclease activity, devoid of or substantially reduced in 5'.fwdarw.3' exonuclease activity, or containing one or mutations in the O-helix that make the enzyme nondiscriminatory for dNTPs and ddNTPs (e.g., a Phe.sup.730 .fwdarw.Tyr.sup.730 mutation of SEQ ID NO: 3) are especially useful for DNA sequencing, DNA labeling, and DNA amplification reactions and cDNA synthesis. Moreover, Thermotoga DNA polymerase mutants containing two or more of these properties are also especially useful for DNA

sequencing, DNA labeling, DNA amplification or cDNA synthesis reactions. As is well known, sequencing reactions (isothermal DNA sequencing and cycle sequencing of DNA) require the use of DNA polymerases. Dideoxy-mediated sequencing involves the use of a chain-termination technique which uses a specific polymer for extension by DNA polymerase, a base-specific chain terminator and the use of polyacrylamide gels to separate the newly synthesized chain-terminated DNA molecules by size so that at least a part of the nucleotide sequence of the original DNA molecule can be determined. Specifically, a DNA molecule is sequenced by using four separate DNA sequence reactions, each of which contains different base-specific terminators. For example, the first reaction will contain a G-specific terminator, the second reaction will contain a T-specific terminator, the third reaction will contain an A-specific terminator, and a fourth reaction may contain a C-specific terminator. Preferred terminator nucleotides include dideoxyribonucleoside triphosphates (ddNTPs) such as ddATP, ddTTP, ddGTP, ddITP and ddCTP. Analogs of dideoxyribonucleoside triphosphates may also be used and are well known in the art.

CLPR:
50. The method of claim 38, wherein said dideoxyribonucleoside triphosphates are selected from the group consisting of ddATP, ddCTP, ddGTP, ddITP, and ddTTP.

CLPR:
66. The method of claim 53, wherein said terminator nucleotides comprise one or more nucleotides selected from the group consisting of ddATP, ddCTP, ddGTP, ddITP, and ddTTP.

CLPR:
74. The kit of claim 69, wherein said dideoxyribonucleoside triphosphates are selected from the group consisting of ddATP, ddCTP, ddGTP, ddITP, and ddTTP.

CLPR:
87. The kit of claim 85, wherein said dideoxyribonucleoside triphosphates are selected from the group consisting of ddATP, ddCTP, ddGTP, ddITP, and ddTTP.

CLPR:
149. The method of claim 136, wherein said terminator nucleotides are selected from the group consisting of ddATP, ddCTP, ddGTP, ddITP, and ddTTP.

---

12. Document ID: US 5939301 A

L8: Entry 12 of 36 File: USPT Aug 17, 1999

US-PAT-NO: 5939301
DOCUMENT-IDENTIFIER: US 5939301 A
TITLE: Cloned DNA polymerases from Thermotoga neapolitana and mutants thereof

AB: The invention relates to a substantially pure thermostable DNA polymerase from Thermotoga neapolitana (Tne) and mutants thereof. The Tne DNA polymerase has a molecular weight of about 100 kilodaltons and is more thermostable than Taq DNA polymerase. The mutant Tne DNA polymerase has at least one mutation selected from the group consisting of (1) a first mutation that substantially reduces or eliminates 3'.fwdarw.5' exonuclease activity of said DNA polymerase; (2) a second mutation that substantially reduces or eliminates 5'.fwdarw.3' exonuclease activity of said DNA polymerase; (3) a third mutation in the O helix of said DNA polymerase resulting in said DNA polymerase becoming non-discriminating against dideoxynucleotides. The present invention also relates to the cloning and expression of the wild type or mutant Tne DNA polymerase in E. coli, to DNA molecules containing the cloned gene, and to host cells which express said genes. The Tne DNA polymerase of the invention may be used in well-known DNA sequencing and amplification reactions.

L8: Entry 12 of 36 File: USPT Aug 17, 1999

DOCUMENT-IDENTIFIER: US 5939301 A
TITLE: Cloned DNA polymerases from Thermotoga neapolitana and mutants thereof

DEPR:
Nucleotide. As used herein "nucleotide" refers to a base-sugar-phosphate combination. Nucleotides are monomeric units of a nucleic acid sequence (DNA and RNA). The term nucleotide includes deoxyribonucleoside triphosphates such as dATP, dCTP, dUTP, dGTP, dTTP, or derivatives thereof. Such derivatives include, for example, dm, [.alpha.S]dATP and 7-deaza-dGTP. The term nucleotide as used herein also refers to dideoxyribonucleoside triphosphates (ddNTPs) and their derivatives. Illustrated examples of dideoxyribonucleoside triphosphates include, but are not limited to, ddATP, ddCTP, ddGTP, ddITP, and ddTTP. According to the present invention, a "nucleotide" may be unlabeled or detectably labeled by well known techniques. Detectable labels include, for example, radioactive isotopes, fluorescent labels, chemiluminescent labels, bioluminescent labels and enzyme labels.

Full | Title | Citation | Front | Review | Classification | Date | Reference KWIC | Draw Desc | Image

---

13. Document ID: US 5912155 A

L8: Entry 13 of 36 File: USPT Jun 15, 1999

US-PAT-NO: 5912155
DOCUMENT-IDENTIFIER: US 5912155 A
TITLE: Cloned DNA polymerases from Thermotoga neapolitana

AB: The invention relates to a substantially pure thermostable DNA polymerase from Thermotoga neapolitana (Tne). The Tne DNA polymerase has a molecular weight of about 100 kilodaltons and is more thermostable than Taq DNA polymerase. The present invention also relates to the cloning and expression of the Tne DNA polymerase in E. coli, to DNA molecules containing the cloned gene, and to host cells which express said genes. The Tne DNA polymerase of the invention may be used in well-known DNA sequencing and amplification reactions.

L8: Entry 13 of 36 File: USPT Jun 15, 1999

DOCUMENT-IDENTIFIER: US 5912155 A
TITLE: Cloned DNA polymerases from Thermotoga neapolitana

DEPR:
Nucleotide. As used herein "nucleotide" refers to a base-sugar-phosphate combination. Nucleotides are monomeric units of a nucleic acid sequence (DNA and RNA). The term nucleotide includes deoxyribonucleoside triphosphates such as dATP, dCTP, dUTP, dGTP, dTTP, or derivatives thereof. Such derivatives include, for example, dm, [.alpha.S]dATP and 7-deaza-dGTP. The term nucleotide as used herein also refers to dideoxyribonucleoside triphosphates (ddNTPs) and their derivatives. Illustrated examples of dideoxyribonucleoside triphosphates include, but are not limited to, ddATP, ddCTP, ddGTP, ddITP, and ddTTP. According to the present invention, a "nucleotide" may be unlabeled or detectably labeled by well known techniques. Detectable labels include, for example, radioactive isotopes, fluorescent labels, chemiluminescent labels, bioluminescent labels and enzyme labels.

Full | Title | Citation | Front | Review | Classification | Date | Reference KWIC | Draw Desc | Image

14. Document ID: US 5871929 A

L8: Entry 14 of 36 File: USPT Feb 16, 1999

US-PAT-NO: 5871929
DOCUMENT-IDENTIFIER: US 5871929 A
TITLE: Suppression of pyrophosphorolysis in DNA sequencing and in other applications involving DNA replication

AB: A method of inhibiting pyrophosphorolysis during DNA chain length elongation is provided. The method comprises including both Mn.sup.++ and Mg.sup.++ in chain-extension and chain-termination reaction mixtures such as those used in DNA sequencing. This method is useful in stabilizing dideoxy-ribonucleoside triphosphate-terminated DNA chains and improving the quality of DNA sequence data obtained via the use of DNA polymerases that do not discriminate against the incorporation of dideoxyribonucleoside triphosphates and other chain terminating agents. Also provided are a reaction mixture and kit for DNA sequencing employing this method.

L8: Entry 14 of 36 File: USPT Feb 16, 1999

DOCUMENT-IDENTIFIER: US 5871929 A
TITLE: Suppression of pyrophosphorolysis in DNA sequencing and in other applications involving DNA replication

DEPR:
Chain-termination reactions typically involve the reaction of hybridized/annealed primer or a hybridized/annealed elongated DNA polymer (formed, for example, by a DNA-extension reaction) with a chain-terminating agent to form a DNA product having a chain-terminating nucleotide incorporated at its 3'-end. The chain-terminating agent is a reagent capable of reacting with a nucleic acid polymer which is hybridized/annealed primer to a nucleic acid template to incorporate at least one nucleotide or other moiety at the 3-end of the polymer, where incorporated nucleotide or other moiety precludes further chain-extension reaction. Typical chain-extending agents include dideoxyribonucleoside triphosphates (ddNTP's) such as ddATP, ddGTP, ddTTP and ddCTP or analogs thereof. Dideoxyribonucleoside analogs which may be suitable for some applications include dideoxyriboinosine triphosphate (ddITP). Non-nucleoside chain-terminating agents--such as acyclovir--may also be employed with the present invention. The chain-extending agents may be labeled or unlabeled, depending on the particular application in which they are being used. Chain-termination reactions are typically carried out in an aqueous solution.

Full | Title | Citation | Front | Review | Classification | Date | Reference KWIC | Draw Desc | Image

15. Document ID: US 5856092 A

L8: Entry 15 of 36 File: USPT Jan 5, 1999

US-PAT-NO: 5856092
DOCUMENT-IDENTIFIER: US 5856092 A
TITLE: Detection of a nucleic acid sequence or a change therein

AB: A method for detecting a specific polynucleotide sequence in a material is disclosed. The method includes exposing said material to an oligonucleotide primer having a sequence complementary to part of said specific polynucleotide sequence wherein said primer binds to part of said polynucleotide sequence when present in said material. Primer bound to the polynucleotide sequence is extended wherein any extended primer includes a detectable element and/or a separation element. Any extended primer is then separated into a fraction wherein said fraction does not have detectable element not included in said extended primer. One then determines whether any extended primer is present in said fraction by assaying said fraction for said extended primer wherein the presence of said extended primer is indicative of the presence of the specific polynucleotide sequence in said material and the absence of said extended primer is indicative of the presence of the specific polynucleotide sequences in said material and the absence of said extended primer in said fraction is indicative of the absence of the specific polynucleotide sequence in said material.

L8: Entry 15 of 36 File: USPT Jan 5, 1999

DOCUMENT-IDENTIFIER: US 5856092 A
TITLE: Detection of a nucleic acid sequence or a change therein

BSPR:
Advantageously, for the first, second, seventh and eighth embodiments extending the primer comprises using at least one nucleotide selected from the group consisting of dATP, dCTP, dGTP, dTTP, dUTP, dITP, ddATP, ddCTP, ddGTP, ddITP and ddTTP. Typically two to four different nucleotides, and especially four different nucleotides are used.

BSPR:
Generally, for the third, fifth, ninth and eleventh embodiments extending the primer comprises using one nucleotide selected from the group consisting of dATP, dCTP, dGTP, dTTP, dUTP, dITP, ddATP, ddCTP, ddGTP, ddITP and ddTTP. Typically one nucleotide selected from the group consisting of ddATP, ddCTP, ddGTP, ddITP and ddTTP is used.

BSPR:
Typically, for the fourth, sixth, tenth and twelfth embodiments extending the primer comprises using at least one nucleotide selected from the group consisting of dATP, dCTP, dGTP, dTTP, dUTP, dITP, ddATP, ddCTP, ddGTP, ddITP and ddTTP. Generally at least one nucleotide selected from the group consisting of ddATP, ddCTP, ddGTP, ddITP and ddTTP is used.

BSPV:
ddNTP--all four dideoxyribonucleotides that is ddATP, ddCTP, ddGTP and ddTTP as well as ddITP

BSPV:
c) at least one nucleotide selected from the group consisting of dATP, dCTP, dGTP, dTTP, dUTP, dITP, ddATP, ddCTP, ddGTP, ddITP and ddTTP; and

BSPV:
c) at least one nucleotide selected from the group consisting of dATP, dCTP, dGTP, dTTP, dUTP, dITP, ddATP, ddCTP, ddGTP, ddITP and ddTTP; and

BSPV:
c) one nucleotide selected from the group consisting of dATP, dCTP, dGTP, dTTP, dUTP, dITP, ddITP, ddATP, ddCTP, ddGTP and ddTTP with the proviso that said nucleotide is selected whereby bound primer is only extended up to and including said specific nucleotide or base when said specific nucleotide or base is at the particular position in the specific polynucleotide sequence; and

BSPV:
c) at least one nucleotide selected from the group consisting of dATP, dCTP, dGTP, dTTP, dUTP, dITP, ddITP, ddATP, ddCTP, ddGTP and ddTTP with the proviso that said nucleotide is selected whereby bound primer is only extended up to and including said specific nucleotide or base when said specific nucleotide or base

is at the particular position in the specific polynucleotide sequence; and

BSPV:
c) one nucleotide selected from the group consisting of dATP, dCTP, dGTP, dTTP, dUTP, dITP, ddITP, ddATP, ddCTP, ddGTP and ddTTP with the provision that said nucleotide is selected whereby bound primer is only extended up to and including said specific nucleotide or base when said specific nucleotide or base is at the particular position in the specific polynucleotide sequence with the proviso that the intervening sequence cannot be one where bases or nucleotides complementary to the intervening sequence and which are incorporated into the extended primer cause incorporation of an interfering detectable and/or separation element; and

BSPV:
c) at least one nucleotide selected from the group consisting of dATP, dCTP, dGTP, dTTP, dUTP, dITP, ddITP, ddATP, ddCTP, ddGTP and ddTTP with the proviso that said nucleotide is selected whereby bound primer is only extended up to and including said specific nucleotide or base when said specific nucleotide or base is at the particular position in the specific polynucleotide sequence with the proviso that the intervening sequences cannot be ones where bases or nucleotides complementary to the intervening sequences and which are incorporated into the extended primer(s) cause incorporation of an interfering detectable and/or separation element(s); and

CLPR:
11. The method of any one of claims 1, 5, or 6 wherein said extending step comprises using four different nucleotides selected from the group consisting of dATP, dCTP, dGTP, dTTP, dUTP, dITP, ddATP, ddCTP, ddGTP, ddITP, and ddTTP, and wherein said nucleotides incorporate at least one element selected from the group consisting of a detectable element and a separation element.

CLPR:
15. The method of any one of claims 1, 5, or 6 wherein said extending step comprises using one nucleotide sequence selected from the group consisting of ddITP, ddATP, ddCTP, ddGTP, and ddTTP, and wherein said nucleotide incorporates at least one element selected from the group consisting of a detectable element and a separation element.

---

16. Document ID: US 5830644 A

L8: Entry 16 of 36 File: USPT Nov 3, 1998

US-PAT-NO: 5830644
DOCUMENT-IDENTIFIER: US 5830644 A
TITLE: Method for screening for agents which increase telomerase activity in a cell

AB: Method and compositions are provided for the determination of telomere length and telomerase activity, as well as the ability to increase or decrease telomerase activity in the treatment of proliferative diseases. Particularly, primers are elongated under conditions which minimize interference from other genomic sequences, so as to obtain accurate determinations of telomeric length or telomerase activity. In addition, compositions are provided for intracellular inhibition of telomerase activity and means are shown for slowing or reversing the loss of telomeric repeats in aging cells.

L8: Entry 16 of 36 File: USPT Nov 3, 1998

DOCUMENT-IDENTIFIER: US 5830644 A
TITLE: Method for screening for agents which increase telomerase activity in a cell

DRPR:
FIG. 20A and B show the effect of nucleoside triphosphate analogs on pausing patterns and processivity of telomerase in vitro. Specifically, FIG. 20A shows telomerase reactions in the presence or absence of. the indicated nucleoside triphosphate analogs. Unlabeled TTP competitor was also analyzed as a control, with and without primer in the reaction mix. Products were then analyzed on a denaturing polyacrylamide gel. FIG. 20B shows standard telomerase reactions were performed in the presence of ddGTP (lanes 4-6), ddITP (lanes 7-9), or DMSO (lane 1). DMSO was the solvent for ddGTP and at the highest concentration tested (1%) showed no effect on the reactions compared with control reactions run without analog or DMSO (control lanes 2-3). Products were analyzed on a denaturing polyacrylamide gel.

DEPR:
We analyzed the effects of various nucleoside analogs, which are chain-terminating inhibitors of retroviral reverse transcriptases, on Tetrahymena thermophila telomerase activity in vitro, and on telomere length and maintenance, cell division and conjugation of Tetrahymena cells in vivo. In vitro assays of telomerase activity showed that arabinofuranyl-guanosine triphosphate (Ara-GTP) and ddGTP were both very efficient inhibitors of incorporation of labeled nucleotides into telomeric DNA repeats, even at low inhibitor concentrations, while azidothymidine triphosphate .(AZT-TP), dideoxyinosine triphosphate (ddITP) or ddTTP were less efficient inhibitors of incorporation. All of these nucleoside triphosphate analogs, however, produced analog-specific alterations of the normal banding patterns seen upon gel electrophoresis of the synthesis products of telomerase, suggesting that the competitive and/or chain terminating action differed at different positions along the RNA template.

DEPR:
We also tested the effects of dideoxynucleoside triphosphates (ddNTPs) on the telomerase reaction. As shown previously for telomerase [Greider 43 Cell, 405, 1985], and as is the case for many other reverse trancriptases, ddNTPs are recognized by the enzyme and incorporated, causing chain termination with a subsequent shift in banding patterns and reduction of the average product length. Consistent with previous qualitative analyses of Tetrahymena and human telomerases [Greider 43 Cell, 405, 1985; Morin 59 Cell, 521, 1989], ddGTP and ddTTP each inhibited the incorporation of labeled .sup.32 P-NTP into elongation products (FIG. 19D and E). ddGTP was a much more efficient inhibitor than ddTTP: under these reaction conditions 50% inhibition occurred at <0.1 and 5 gM ddGTP and ddTTP respectively. As observed previously for Tetrahymena telomerase [Greider 43 Cell, 405, 1985], no significant effects were seen with either ddCTP or ddATP. In addition, ddITP inhibited telomerase (FIG. 19E), although less efficiently than ddGTP, with 50% inhibition occurring at 3 gM ddITP.

DEPR:
The size distribution of labeled products was then analyzed by denaturing polyacrylamide gel electrophoresis. Consistent with the expectation for a chain-terminator, the proportion of longer telomerase products was decreased in the presence of AZT compared with cold TTP competitor controls (FIG. 20A; compare lanes 1 and 2 with lanes 3 to 5), and in the presence of Ara-G (FIG. 20A; lanes 7 and 8). Average product length also decreased in the presence of Ara-GTP, ddGTP and ddITP (FIG. 20A and B). In addition, each nucleoside triphosphate analog produced distinctive and characteristic patterns of chain termination, as shown by analysis of the shifts in the banding patterns of the elongation products. With AZT-triphosphate, we saw increased relative intensities of the bands corresponding to the incorporation of T residues (copying the A residues at positions 2 and 3 on the template RNA (see FIG. 18A)). This change in banding pattern is consistent with simple chain termination, which is predicted to increase the intensity of bands corresponding to the position of both incorporated T residues. Similar effects were seen with ddTTP. We interpret this to mean that AZT-triphosphate was recognized by the enzyme and incorporated into the correct positions in the growing telomeric sequence, causing chain termination. However it cannot be excluded that the increase in relative intensity of the band corresponding to position 3 on the template, which precedes addition of the second T, is also attributable to pausing caused by competition with TTP and a slower reaction rate with AZT-triphosphate at position 2. Should AZT-triphosphate, or related nucleotide analogs, be incorporated into telomeric DNA where they would not be incorporated by DNA polymerase into other DNA, then

such nucleotide analogs may be used to kill telomerase positive cells by causing them to generate telomeric DNA toxic to the cell, or at least altered in such a way that telomerase-mediated cell immortalization was inhibited.

DEPR:
The results with Ara-GTP were also consistent with incorporation of Ara-G and consequent chain termination (FIG. 20A, lanes 7 and 8). Although there are four positions at which a G residue can be incorporated and therefore at which chain termination could occur, the strongest increase was in the band corresponding to the G residue specified by position 4, in the middle of the telomerase RNA template (see FIG. 18A). With ddGTP, chain termination appeared to occur most efficiently at positions 6 and 5 (FIG. 20B, compare lane 1 with lanes 4 to 6), and with ddITP, at position 5 (lanes 7 to 9).

Full | Title | Citation | Front | Review | Classification | Date | Reference KWIC | Draw Desc | Image

---

17. Document ID: US 5753221 A

L8: Entry 17 of 36 File: USPT May 19, 1998

US-PAT-NO: 5753221
DOCUMENT-IDENTIFIER: US 5753221 A
TITLE: Transformed erythrocytes, process for preparing the same, and their use in pharmaceutical compositions

AB: The invention relates to compositions of erythrocytes that have been modified following hypotonic lysis and resealing by addition of 2',3'-dideoxycytidine-5'-triphosphate (ddCTP) or 3'-azido-3'-deoxythymidine-5'-triphosphate (AZT-TP). These compositions may also contain ATP. Also disclosed are methods of preparing these compositions.

L8: Entry 17 of 36 File: USPT May 19, 1998

DOCUMENT-IDENTIFIER: US 5753221 A
TITLE: Transformed erythrocytes, process for preparing the same, and their use in pharmaceutical compositions

BSPV:
2',3'-dideoxyinosine-5'-triphosphate (ddITP), and/or

BSPV:
2-amino-6-fluoro-2',3'-dideoxyinosine-triphosphate, and/or

DEPR:
Under similar conditions 2',3'-dideoxyadenosine-5'-triphosphate (ddATP), 2',3'-dideoxyguanosine-5'-triphosphate (ddGTP) and 2',3'-dideoxyinosine-5'-triphosphate (ddITP) added to the erythrocyte lysate were stable both in the presence or absence of ATP for at least 2 h at 37.degree. C.

Full | Title | Citation | Front | Review | Classification | Date | Reference KWIC | Draw Desc | Image

---

18. Document ID: US 5695932 A

L8: Entry 18 of 36 File: USPT Dec 9, 1997

US-PAT-NO: 5695932
DOCUMENT-IDENTIFIER: US 5695932 A
TITLE: Telomerase activity assays for diagnosing pathogenic infections

AB: Method and compositions are provided for the determination of telomere length and telomerase activity, as well as the ability to inhibit telomerase activity in the treatment of proliferative diseases. Particularly, primers are elongated under conditions which minimize interference from other genomic sequences, so as to obtain accurate determinations of telomeric length or telomerase activity. In addition, compositions are provided for intracellular inhibition of telomerase activity and means are shown for slowing the loss of telomeric repeats in aging cells.

L8: Entry 18 of 36 File: USPT Dec 9, 1997

DOCUMENT-IDENTIFIER: US 5695932 A
TITLE: Telomerase activity assays for diagnosing pathogenic infections

DRPR:
FIGS. 20A and B show the effect of nucleoside triphosphate analogs on pausing patterns and processivity of telomerase in vitro. Specifically, FIG. 20A shows telomerase reactions in the presence or absence of the indicated nucleoside triphosphate analogs. Unlabeled TTP competitor was also analyzed as a control, with and without primer in the reaction mix. Products were then analyzed on a denaturing polyacrylamide gel. FIG. 20B shows standard telomerase reactions were performed in the presence of ddGTP (lanes 4-6), ddITP (lanes 7-9), or DMSO (lane 1). DMSO was the solvent for ddGTP and at the highest concentration tested (1%) showed no effect on the reactions compared with control reactions run without analog or DMSO (control lanes 2-3). Products were analyzed on a denaturing polyacrylamide gel.

DEPR:
We analyzed the effects of various nucleoside analogs, which are chain-terminating inhibitors of retroviral reverse transcriptases, on Tetrahymena thermophila telomerase activity in vitro, and on telomere length and maintenance, cell division and conjugation of Tetrahymena cells in vivo. In vitro assays of telomerase activity showed that arabinofuranyl-guanosine triphosphate (Ara-GTP) and ddGTP were both very efficient inhibitors of incorporation of labeled nucleotides into telomeric DNA repeats, even at low inhibitor concentrations, while azidothymidine triphosphate (AZT-TP), dideoxyinosine triphosphate (ddITP) or ddTTP were less efficient inhibitors of incorporation. All of these nucleoside triphosphate analogs, however, produced analog-specific alterations of the normal banding patterns seen upon gel electrophoresis of the synthesis products of telomerase, suggesting that the competitive and/or chain terminating action differed at different positions along the RNA template.

DEPR:
We also tested the effects of dideoxynucleoside triphosphates (ddNTPs) on the telomerase reaction. As shown previously for telomerase [Greider 43 Cell, 405, 1985], and as is the case for many other reverse trancriptases, ddNTPs are recognized by the enzyme and incorporated, causing chain termination with a subsequent shift in banding patterns and reduction of the average product length. Consistent with previous qualitative analyses of Tetrahymena and human telomerases [Greider 43 Cell, 405, 1985; Morin 59 Cell, 521, 1989], ddGTP and ddTTP each inhibited the incorporation of labeled .sup.32 P-NTP into elongation products (FIG. 19D and E). ddGTP was a much more efficient inhibitor than ddTTP: under these reaction conditions 50% inhibition occurred at <0.1 and 5 .mu.M ddGTP and ddTTP respectively. As observed previously for Tetrahymena telomerase [Greider 43 Cell, 405, 1985], no significant effects were seen with either ddCTP or ddATP. In addition, ddITP inhibited telomerase (FIG. 19E), although less efficiently than ddGTP, With 50% inhibition occurring at 3 .mu.M ddITP.

DEPR:
The size distribution of labeled products was then analyzed by denaturing polyacrylamide gel electrophoresis. Consistent with the expectation for a chain-terminator, the proportion of longer telomerase products was decreased in the presence of AZT compared with cold TTP competitor controls (FIG. 20A; compare lanes 1 and 2 with lanes 3 to 5), and in the presence of Ara-G (FIG. 20A; lanes 7 and 8). Average product length also decreased in the presence of Ara-GTP, ddGTP and ddITP (FIG. 20A and B). In addition, each nucleoside triphosphate analog produced distinctive and characteristic patterns of chain termination, as shown

by analysis of the shifts in the banding patterns of the elongation products. With AZT-triphosphate, we saw increased relative intensities of the bands corresponding to the incorporation of T residues (copying the A residues at positions 2 and 3 on the template RNA (see FIG. 18A)). This change in banding pattern is consistent with simple chain termination, which is predicted to increase the intensity of bands corresponding to the position of both incorporated T residues. Similar effects were seen with ddTTP. We interpret this to mean that AZT-triphosphate was recognized by the enzyme and incorporated into the correct positions in the growing telomeric sequence, causing chain termination. However it cannot be excluded that the increase in relative intensity of the band corresponding to position 3 on the template, which precedes addition of the second T, is also attributable to pausing caused by competition with TTP and a slower reaction rate with AZT-triphosphate at position 2.

DEPR:
The results with Ara-GTP were also consistent with incorporation of Ara-G and consequent chain termination (FIG. 20A, lanes 7 and 8). Although there are four positions at which a G residue can be incorporated and therefore at which chain termination could occur, the strongest increase was in the band corresponding to the G residue specified by position 4, in the middle of the telomerase RNA template (see FIG. 18A). With ddGTP, chain termination appeared to occur most efficiently at positions 6 and 5 (FIG. 20B, compare lane 1 with lanes 4 to 6), and with ddITP, at position 5 (lanes 7 to 9).

19. Document ID: US 5691141 A

L8: Entry 19 of 36 File: USPT Nov 25, 1997

US-PAT-NO: 5691141
DOCUMENT-IDENTIFIER: US 5691141 A
TITLE: DNA sequencing by mass spectrometry

AB: The invention describes a new method to sequence DNA. The improvements over the existing DNA sequencing technologies are high speed, high throughput, no electrophoresis and gel reading artifacts due to the complete absence of an electrophoretic step, and no costly reagents involving various substitutions with stable isotopes. The invention utilizes the Sanger sequencing strategy and assembles the sequence information by analysis of the nested fragments obtained by base-specific chain termination via their different molecular masses using mass spectrometry, as for example, MALDI or ES mass spectrometry. A further increase in throughput can be obtained by introducing mass-modifications in the oligonucleotide primer, chain-terminating nucleoside triphosphates and/or in the chain-elongating nucleoside triphosphates, as well as using integrated tag sequences which allow multiplexing by hybridization of tag specific probes with mass differentiated molecular weights.

L8: Entry 19 of 36 File: USPT Nov 25, 1997

DOCUMENT-IDENTIFIER: US 5691141 A
TITLE: DNA sequencing by mass spectrometry

CLPR:
3. A kit of claim 1, wherein the set of chain-terminating nucleotides is comprised of the following nucleotides: dideoxyadenosine triphosphate (ddATP), dideoxythymidine triphosphate (ddTTP), dideoxyguanosine triphosphate (ddGTP), dideoxycytidine triphosphate (ddCTP), 7-deaza dideoxyguanosine triphosphate (c.sup.7 ddGTP), 7-deaza dideoxyadenosine triphosphate (c.sup.7 ddATP), 7-deaza dideoxyinosine triphosphate (c.sup.7 ddITP).

CLPR:
17. A kit of claim 15, wherein the set of chain-terminating nucleotides is comprised of the following nucleotides: dideoxyadenosine triphosphate (ddATP), dideoxythymidine triphosphate (ddTTP), dideoxyguanosine triphosphate (ddGTP), dideoxycytidine triphosphate (ddCTP), 7-deazadideoxyguanosine triphosphate (c.sup.7 ddGTP), 7-deazadideoxyadenosine triphosphate (c.sup.7 ddATP), 7-deazadideoxyinosine triphosphate (c.sup.7 ddITP).

20. Document ID: US 5674716 A

L8: Entry 20 of 36 File: USPT Oct 7, 1997

US-PAT-NO: 5674716
DOCUMENT-IDENTIFIER: US 5674716 A
TITLE: DNA sequencing

AB: A method for sequencing a strand of DNA, including the steps of: providing the strand of DNA; annealing the strand with a primer able to hybridize to the strand to give an annealed mixture; incubating the mixture with a deoxyribonucleoside triphosphate, a DNA polymerase, and a chain terminating agent under conditions in which the polymerase causes the primer to be elongated to form a series of DNA products differing in length of the elongated primer, each DNA product having a chain terminating agent at its elongated end; the number of each DNA product being approximately the same for substantially all DNA products differing in length from 1 to 20 bases.

L8: Entry 20 of 36 File: USPT Oct 7, 1997

DOCUMENT-IDENTIFIER: US 5674716 A
TITLE: DNA sequencing

DEPR:
The DNA polymerases of this invention do not discriminate significantly between dideoxynucleoside analogs and deoxynucleosides along the length of the DNA template. That is, in the presence of manganese or iron these polymerases are unable to discriminate between a nucleotide that has a 3' hydroxyl group versus one that does not (i.e., has two hydrogens at the 3' position of the ribose). However, these polymerases do discriminate against modifications at other positions on the nucleosides, even in the presence of manganese or iron. For example, the polymerases do discriminate against some dideoxynucleoside analogs which have fluorescent groups attached compared to deoxynucleosides. However, the polymerase do not discriminate to a different extent at neighboring, or nearby nucleotides, on the basis of the presence or absence of the modification to the dideoxynucleoside. Thus, while they discriminate strongly against these analogs, requiring higher concentrations for a DNA sequencing reaction compared to unmodified dideoxynucleosides, the intensity of nearby bands will still be uniform. For example, there is a 10 fold discrimination against dideoxy ITP (ddITP), compared to dideoxy GTP (ddGTP), in the presence of Mn. However, all the bands produced in a sequencing reaction are of equal intensity with ddITP since there is no differential discrimination along the length of the DNA template.

☐ 21. Document ID: US 5645986 A

L8: Entry 21 of 36 File: USPT Jul 8, 1997

US-PAT-NO: 5645986
DOCUMENT-IDENTIFIER: US 5645986 A
TITLE: Therapy and diagnosis of conditions related to telomere length and/or telomerase activity

AB: Method and compositions are provided for the determination of telomere length and telomerase activity, as well as the ability to increase or decrease telomerase activity in the treatment of proliferative diseases. Particularly, primers are elongated under conditions which minimize interference from other genomic sequences, so as to obtain accurate determinations of telomeric length or telomerase activity. In addition, compositions are provided for intracellular inhibition of telomerase activity and means are shown for slowing or reversing the loss of telomeric repeats in aging cells.

L8: Entry 21 of 36 File: USPT Jul 8, 1997

DOCUMENT-IDENTIFIER: US 5645986 A
TITLE: Therapy and diagnosis of conditions related to telomere length and/or telomerase activity

DRPR:
FIGS. 20A and B show the effect of nucleoside triphosphate analogs on pausing patterns and processivity of telomerase in vitro. Specifically, FIG. 20A shows telomerase reactions in the presence or absence of the indicated nucleoside triphosphate analogs. Unlabeled TTP competitor was also analyzed as a control, with and without primer in the reaction mix. Products were then analyzed on a denaturing polyacrylamide gel. FIG. 20B shows standard telomerase reactions were performed in the presence of ddGTP (lanes 4-6), ddITP (lanes 7-9), or DMSO (lane 1). DMSO was the solvent for ddGTP and at the highest concentration tested (1%) showed no effect on the reactions compared with control reactions run without analog or DMSO (control lanes 2-3). Products were analyzed on a denaturing

polyacrylamide gel.

DEPR:
We analyzed the effects of various nucleoside analogs, which are chain-terminating inhibitors of retroviral reverse transcriptases, on Tetrahymena thermophila telomerase activity in vitro, and on telomere length and maintenance, cell division and conjugation of Tetrahymena cells in vivo. In vitro assays of telomerase activity showed that arabinofuranyl-guanosine triphosphate (Ara-GTP) and ddGTP were both very efficient inhibitors of incorporation of labeled nucleotides into telomeric DNA repeats, even at low inhibitor concentrations, while azidothymidine triphosphate (AZT-TP), dideoxyinosine triphosphate (ddITP) or ddTTP were less efficient inhibitors of incorporation. All of these nucleoside triphosphate analogs, however, produced analog-specific alterations of the normal banding patterns seen upon gel electrophoresis of the synthesis products of telomerase, suggesting that the competitive and/or chain terminating action differed at different positions along the RNA template.

DEPR:
We also tested the effects of dideoxynucleoside triphosphates (ddNTPs) on the telomerase reaction. As shown previously for telomerase [Greider 43 Cell, 405, 1985], and as is the case for many other reverse trancriptases, ddNTPs are recognized by the enzyme and incorporated, causing chain termination with a subsequent shift in banding patterns and reduction of the average product length. Consistent with previous qualitative analyses of Tetrahymena and human telomerases [Greider 43 Cell, 405, 1985; Morin 59 Cell, 521, 1989], ddGTP and ddTTP each inhibited the incorporation of labeled .sup.32 P-NTP into elongation products (FIG. 19D and E). ddGTP was a much more efficient inhibitor than ddTTP: under these reaction conditions 50% inhibition occurred at <0.1 and 5 .mu.M ddGTP and ddTTP respectively. As observed previously for Tetrahymena telomerase [Greider 43 Cell, 405, 1985], no significant effects were seen with either ddCTP or ddATP. In addition, ddITP inhibited telomerase (FIG. 19E), although less efficiently than ddGTP, with 50% inhibition occurring at 3 .mu.M ddITP.

DEPR:
The size distribution of labeled products was then analyzed by denaturing polyacrylamide gel electrophoresis. Consistent with the expectation for a chain-terminator, the proportion of longer telomerase products was decreased in the presence of AZT compared with cold TTP competitor controls (FIG. 20A; compare lanes 1 and 2 with lanes 3 to 5), and in the presence of Ara-G (FIG. 20A; lanes 7 and 8). Average product length also decreased in the presence of Ara-GTP, ddGTP and ddITP (FIG. 20A and B). In addition, each nucleoside triphosphate analog produced distinctive and characteristic patterns of chain termination, as shown by analysis of the shifts in the banding patterns of the elongation products. With AZT-triphosphate, we saw increased relative intensities of the bands corresponding to the incorporation of T residues (copying the A residues at positions 2 and 3 on the template RNA (see FIG. 18A)). This change in banding pattern is consistent with simple chain termination, which is predicted to increase the intensity of bands corresponding to the position of both incorporated T residues. Similar effects were seen with ddTTP. We interpret this to mean that AZT-triphosphate was recognized by the enzyme and incorporated into the correct positions in the growing telomeric sequence, causing chain termination. However it cannot be excluded that the increase in relative intensity of the band corresponding to position 3 on the template, which precedes addition of the second T, is also attributable to pausing caused by competition with TTP and a slower reaction rate with AZT-triphosphate at position 2. Should AZT-triphosphate, or related nucleotide analogs, be incorporated into telomeric DNA where they would not be incorporated by DNA polymerase into other DNA, then such nucleotide analogs may be used to kill telomerase positive cells by causing them to generate telomeric DNA toxic to the cell, or at least altered in such a way that telomerase-mediated cell immortalization was inhibited.

DEPR:
The results with Ara-GTP were also consistent with incorporation of Ara-G and consequent chain termination (FIG. 20A, lanes 7 and 8). Although there are four positions at which a G residue can be incorporated and therefore at which chain termination could occur, the strongest increase was in the band corresponding to the G residue specified by position 4, in the middle of the telomerase RNA template (see FIG. 18A). With ddGTP, chain termination appeared to occur most efficiently at positions 6 and 5 (FIG. 20B, compare lane 1 with lanes 4 to 6),

and with ddITP, at position 5 (lanes 7 to 9).

Full | Title | Citation | Front | Review | Classification | Date | Reference KWIC | Draw Desc | Image

---

22. Document ID: US 5639608 A

L8: Entry 22 of 36 File: USPT Jun 17, 1997

US-PAT-NO: 5639608
DOCUMENT-IDENTIFIER: US 5639608 A
TITLE: Method for sequencing DNA using a T7-type DNA polymerase and short oligonucleotide primers

AB: This invention relates to methods for determining the nucleotide base sequence of a deoxyribose nucleic acid molecule comprising the steps of incubating the nucleic acid molecule with an oligonucleotide primer of 5 to 8 bases in length, a plurality of deoxynucleoside triphosphates, at least one chain terminating agent, and a T7-type DNA polymerase having less than 500 units of exonuclease activity under conditions in which the primer is extended until the chain terminating agent is incorporated, and separating the products of the incubating step according to size, whereby at least a part of the nucleotide base sequence of the nucleic acid molecule can be determined.

L8: Entry 22 of 36 File: USPT Jun 17, 1997

DOCUMENT-IDENTIFIER: US 5639608 A
TITLE: Method for sequencing DNA using a T7-type DNA polymerase and short oligonucleotide primers

DEPR:
Modified T7 DNA polymerase efficiently utilizes dITP or deaza-GTP in place of dGTP. dITP is substituted for dGTP in both the pulse and chase mixes at a concentration two to five times that at which dGTP is used. In the ddG chase mix ddGTP is still used (not ddITP).

Full | Title | Citation | Front | Review | Classification | Date | Reference KWIC | Draw Desc | Image

---

23. Document ID: US H001531 H

L8: Entry 23 of 36 File: USPT May 7, 1996

US-PAT-NO: H001531
DOCUMENT-IDENTIFIER: US H001531 H
TITLE: Thermophilic DNA polymerase

AB: The invention relates to a substantially pure thermostable DNA polymerase. Preferably, the DNA polymerase has a molecular weight of about 95 kilodaltons and is more thermostable than Taq DNA polymerase. The present invention also relates to cloning and expression of the DNA polymerase in E. coli, to DNA molecules containing the cloned gene, and to host cells which express said genes.

L8: Entry 23 of 36 File: USPT May 7, 1996

DOCUMENT-IDENTIFIER: US H001531 H
TITLE: Thermophilic DNA polymerase

BSPR:
Nucleotide. As used herein "nucleotide" refers to a base-sugar-phosphate combination. Nucleotides are monomeric units of a nucleic acid sequence (DNA and RNA). The term nucleotide includes deoxyribonucleoside triphosphates such as dATP, dCTP, dUTP, dGTP, dTTP, or derivatives thereof. Such derivatives include, for example, dITP, .alpha.SdATP and 7-deaza-dGTP. The term nucleotide as used herein also refers to dideoxyribonucleoside triphosphates (ddNTPs) and their derivatives. Illustrated examples of dideoxyribonucleoside triphosphates include, but are not limited to, ddATP, ddCTP, ddGTP, ddITP, and ddTTP. According to the present invention, a "nucleotide" may be unlabeled or detectably labeled by well known techniques. Detectable labels include, for example, radioactive isotopes, fluorescent labels, chemiluminescent labels, bioluminescent labels and enzyme labels.

---

24. Document ID: US 5409811 A

L8: Entry 24 of 36 File: USPT Apr 25, 1995

US-PAT-NO: 5409811
DOCUMENT-IDENTIFIER: US 5409811 A
TITLE: DNA sequencing

AB: A method for sequencing a strand of DNA, including the steps off: providing the strand of DNA; annealing the strand with a primer able to hybridize to the strand to give an annealed mixture; incubating the mixture with four deoxyribonucleoside triphosphates, a DNA polymerase, and at least three deoxyribonucleoside triphosphates in different amounts, under conditions in favoring primer extension to form nucleic acid fragments complementory to the DNA to be sequenced; labelling the nucleic and fragments; separating them and determining the position of the deoxyribonucleoside triphosphates by differences in the intensity of the labels, thereby to determine the DNA sequence.

L8: Entry 24 of 36 File: USPT Apr 25, 1995

DOCUMENT-IDENTIFIER: US 5409811 A
TITLE: DNA sequencing

DEPR:
The DNA polymerases of this invention do not discriminate significantly between dideoxynucleoside analogs and deoxynucleosides along the length of the DNA template. That is, in the presence of manganese or iron these polymerases are unable to discriminate between a nucleotide that has a 3' hydroxyl group versus one that does not (i.e., has two hydrogens at the 3' position of the ribose). However, these polymerases do discriminate against modifications at other positions on the nucleosides, even in the presence of manganese or iron. For example, the polymerases do discriminate against some dideoxynucleoside analogs which have fluorescent groups attached compared to deoxynucleosides. However, the polymerase do not discriminate to a different extent at neighboring, or nearby nucleotides, on the basis of the presence or absence of the modification to the dideoxynucleoside. Thus, while they discriminate strongly against these analogs, requiring higher concentrations for a DNA sequencing reaction compared to unmodified dideoxynucleosides, the intensity of nearby bands will still be uniform. For example, there is a 10 fold discrimination against dideoxy ITP (ddITP), compared to dideoxy GTP (ddGTP), in the presence of Mn. However, all the bands produced in a sequencing reaction are of equal intensity with ddITP since there is no differential discrimination along the length of the DNA template.

Full | Title | Citation | Front | Review | Classification | Date | Reference | KWIC | Draw Desc | Image

---

☐ 25. Document ID: US 5366860 A

L8: Entry 25 of 36 File: USPT Nov 22, 1994

US-PAT-NO: 5366860
DOCUMENT-IDENTIFIER: US 5366860 A
TITLE: Spectrally resolvable rhodamine dyes for nucleic acid sequence determination

AB: A spectrally resolvable set of rhodamine dyes are provided for use in the chain termination method of nucleic acid sequencing. A different rhodamine dye from the group consisting of tetramethylrhodamine, rhodamine X, rhodamine 6G, and rhodamine 110 is attached to the base of each of the dideoxynucleotides used in the sequencing method by way of an alkynylamino linker. Preferably, the labeled dideoxynucleotides are incorporated into the growing DNA chains by Taq DNA polymerase.

L8: Entry 25 of 36 File: USPT Nov 22, 1994

DOCUMENT-IDENTIFIER: US 5366860 A
TITLE: Spectrally resolvable rhodamine dyes for nucleic acid sequence determination

DEPC:
Preparation of R110-labeled 2',3'-dideoxyinosine triphosphate (ddG-5R110)

Full | Title | Citation | Front | Review | Classification | Date | Reference | KWIC | Draw Desc | Image

---

☐ 26. Document ID: US 5266466 A

L8: Entry 26 of 36 File: USPT Nov 30, 1993

US-PAT-NO: 5266466
DOCUMENT-IDENTIFIER: US 5266466 A
TITLE: Method of using T7 DNA polymerase to label the 3' end of a DNA molecule

AB: This invention relates to T7-type DNA polymerases and methods for using them.

L8: Entry 26 of 36 File: USPT Nov 30, 1993

DOCUMENT-IDENTIFIER: US 5266466 A
TITLE: Method of using T7 DNA polymerase to label the 3' end of a DNA molecule

DEPR:
Modified T7 DNA polymerase efficiently utilizes dITP or deaza-GTP in place of dGTP. dITP is substituted for dGTP in both the pulse and chase mixes at a concentration two to five times that at which dGTP is used. In the ddG chase mix ddGTP is still used (not ddITP).

Full | Title | Citation | Front | Review | Classification | Date | Reference | KWIC | Draw Desc | Image

---

27. Document ID: US 5187085 A

L8: Entry 27 of 36 File: USPT Feb 16, 1993

US-PAT-NO: 5187085
DOCUMENT-IDENTIFIER: US 5187085 A
TITLE: Nucleic acid sequence analysis with nucleoside-5'-O-(1-thiotriphosphates)

AB: A chain-termination method of nucleic acid sequence determination is provided wherein nucleoside triphosphate precursors are replaced with their 1-thiotriphosphate analogs in the polymerization step. This substitution results in more uniform bands of electrophoretically separated DNA fragments which, in turn, results in more accurate base determination.

L8: Entry 27 of 36 File: USPT Feb 16, 1993

DOCUMENT-IDENTIFIER: US 5187085 A
TITLE: Nucleic acid sequence analysis with nucleoside-5'-O-(1-thiotriphosphates)

DEPC:
Preparation of R110-labeled 2',3'-dideoxyinosine triphosphate (ddG-5R110)

Full | Title | Citation | Front | Review | Classification | Date | Reference | KWIC | Draw Desc | Image

---

28. Document ID: US 5173411 A

L8: Entry 28 of 36 File: USPT Dec 22, 1992

US-PAT-NO: 5173411
DOCUMENT-IDENTIFIER: US 5173411 A
TITLE: Method for determining the nucleotide base sequence of a DNA molecule

AB: This invention relates to T7-type DNA polymerases and methods for using them.

L8: Entry 28 of 36 File: USPT Dec 22, 1992

DOCUMENT-IDENTIFIER: US 5173411 A
TITLE: Method for determining the nucleotide base sequence of a DNA molecule

DEPR:
Modified T7 DNA polymerase efficiently utilizes dITP or deaza-GTP in place of dGTP. dITP is substituted for dGTP in both the pulsed chase mixes at a concentration two to five times that at which dGTP is used. In the ddG chase mix ddGTP is still used (not ddITP).

Full | Title | Citation | Front | Review | Classification | Date | Reference KWIC | Draw Desc | Image

---

29. Document ID: US 5145776 A

L8: Entry 29 of 36 File: USPT Sep 8, 1992

US-PAT-NO: 5145776
DOCUMENT-IDENTIFIER: US 5145776 A
TITLE: Method of using T7 DNA polymerase to mutagenize and fill-in DNA

AB: Methods for producing blunt-ended double stranded DNA, for labelling the 3'-end of a DNA fragment, and for in vitro mutagenesis of a DNA fragment. A processive DNA polymerase is used in each method.

L8: Entry 29 of 36 File: USPT Sep 8, 1992

DOCUMENT-IDENTIFIER: US 5145776 A
TITLE: Method of using T7 DNA polymerase to mutagenize and fill-in DNA

DEPR:
Modified T7 DNA polymerase efficiently utilizes dITP or deaza-GTP in place of dGTP. dITP is substituted for dGTP in both the pulse and chase mixes at a concentration two to five times that at which dGTP is used. In the ddG chase mix ddGTP is still used (not ddITP).

Full | Title | Citation | Front | Review | Classification | Date | Reference KWIC | Draw Desc | Image

---

30. Document ID: US 5122345 A

L8: Entry 30 of 36 File: USPT Jun 16, 1992

US-PAT-NO: 5122345
DOCUMENT-IDENTIFIER: US 5122345 A
TITLE: DNA Sequencing apparatus

AB: This invention was made with government support including a grant from the U.S. Public Health Service, contract number AI-06045. The U.S. government has certain rights in the invention., An automated DNA sequencing apparatus having a reactor for providing at least two series of DNA products formed from a single primer and a DNA strand, each DNA product of a series differing in molecular weight and having a chain terminating agent at one end; separating means for separating the DNA products to form a series bands, the intensity of substantially all nearby bands in a different series being different, band reading means for determining the position an

L8: Entry 30 of 36 File: USPT Jun 16, 1992

DOCUMENT-IDENTIFIER: US 5122345 A
TITLE: DNA Sequencing apparatus

DEPR:
The DNA polymerases of this invention do not discriminate significantly between dideoxynucleoside analogs and deoxynucleosides along the length of the DNA template. That is, in the presence of manganese or iron these polymerases are unable to discriminate between a nucleotide that has a 3' hydroxyl group versus one that does not (i.e., has two hydrogens at the 3' position of the ribose). However, these polymerases do discriminate against modifications at other positions on the nucleosides, even in the presence of manganese or iron. For example, the polymerases do discriminate against some dideoxynucleoside analogs which have fluorescent groups attached compared to deoxynucleosides. However, the polymerase do not discriminate to a different extent at neighboring, or nearby nucleotides, on the basis of the presence or absence of the modification to the dideoxynucleoside. Thus, while they discriminate strongly against these analogs, requiring higher concentrations for a DNA sequencing reaction compared to unmodified dideoxynucleosides, the intensity of nearby bands will still be uniform. For example, there is a 10 fold discrimination against dideoxy ITP (ddITP), compared to dideoxy GTP (ddGTP), in the presence of Mn. However all the bands produced in a sequencing reaction are of equal intensity with ddITP since there is no differential discrimination along the length of the DNA template.

Full | Title | Citation | Front | Review | Classification | Date | Reference | KWIC | Draw Desc | Image

---

31. Document ID: US 4994372 A

L8: Entry 31 of 36 File: USPT Feb 19, 1991

US-PAT-NO: 4994372
DOCUMENT-IDENTIFIER: US 4994372 A
TITLE: DNA sequencing

AB: This invention relates to processive DNA polymerases and methods for using them.

L8: Entry 31 of 36 File: USPT Feb 19, 1991

DOCUMENT-IDENTIFIER: US 4994372 A
TITLE: DNA sequencing

DEPR:
Modified T7 DNA polymerase efficiently utilizes dITP or deaza-GTP in place of dGTP. dITP is substituted for dGTP in both the pulse and chase mixes at a concentration two to five times that at which dGTP is used. In the ddG chase mix ddGTP is still used (not ddITP).

32. Document ID: US 4962020 A

L8: Entry 32 of 36 File: USPT Oct 9, 1990

US-PAT-NO: 4962020
DOCUMENT-IDENTIFIER: US 4962020 A
TITLE: DNA sequencing

AB: A method for sequencing a strand of DNA, including the steps of: providing the strand of DNA; annealing the strand with a primer able to hybridize to the strand to give an annealed mixture; incubating the mixture with a deoxyribonucleoside triphosphate, a DNA polymerase, and a chain terminating agent under conditions in which the polymerase causes the primer to be elongated to form a series of DNA products differing in length of the elongated primer, each DNA product having a chain terminating agent at its elongated end; the number of each DNA product being approximately the same for substantially all DNA products differing in length from 1 to 20 bases.

L8: Entry 32 of 36 File: USPT Oct 9, 1990

DOCUMENT-IDENTIFIER: US 4962020 A
TITLE: DNA sequencing

DEPR:
The DNA polymerases of this invention do not discriminate significantly between dideoxynucleoside analogs and deoxynucleosides along the length of the DNA template. That is, in the presence of manganese or iron these polymerases are unable to discriminate between a nucleotide that has a 3' hydroxyl group versus one that does not (i.e., has two hydrogens at the 3' position of the ribose). However, these polymerases do discriminate against modifications at other positions on the nucleosides, even in the presence of manganese or iron. For example, the polymerases do discriminate against some dideoxynucleoside analogs which have fluorescent groups attached compared to deoxynucleosides. However, the polymerase do not discriminate to a different extent at neighboring, or nearby nucleotides, on the basis of the presence or absence of the modification to the dideoxynucleoside. Thus, while they discriminate strongly against these analogs, requiring higher concentrations for a DNA sequencing reaction compared to unmodified dideoxynucleosides, the intensity of nearby bands will still be uniform. For example, there is a 10 fold discrimination against dideoxy ITP (ddITP), compared to dideoxy GTP (ddGTP), in the presence of Mn However, all the bands produced in a sequencing reaction are of equal intensity with ddITP since there is no differential discrimination along the length of the DNA template.

Full | Title | Citation | Front | Review | Classification | Date | Reference KWIC | Draw Desc | Image

33. Document ID: US 4946786 A

L8: Entry 33 of 36 File: USPT Aug 7, 1990

US-PAT-NO: 4946786
DOCUMENT-IDENTIFIER: US 4946786 A
TITLE: T7 DNA polymerase

AB: 1Method for production of a composition consisting essentially of a T7-type DNA polymerase and thioredoxin. The method includes culturing a cell containing plasmid DNA encoding a T7-type DNA polymerase to express the T7-type DNA polymerase from the plasmid DNA, and purifying the T7-type DNA polymerase expressed from the cell to reduce the exonuclease activity associated with the T7-type DNA polymerase compared to the level of exonuclease activity associated with a corresponding naturally-occurring T7-type DNA polymerase.

L8: Entry 33 of 36 File: USPT Aug 7, 1990

DOCUMENT-IDENTIFIER: US 4946786 A
TITLE: T7 DNA polymerase

DEPR:
Modified T7 DNA polymerase efficiently utilizes dITP or deaza-GTP in place of dGTP. dITP is substituted for dGTP in both the pulse and chase mixes at a concentration two to five times that at which dGTP is used. In the ddG chase mix ddGTP is still used (not ddITP).

Full | Title | Citation | Front | Review | Classification | Date | Reference | KWIC | Draw Desc | Image

---

☐ 34. Document ID: US 4942130 A

L8: Entry 34 of 36 File: USPT Jul 17, 1990

US-PAT-NO: 4942130
DOCUMENT-IDENTIFIER: US 4942130 A
TITLE: T7 DNA polymerase

AB: No data.

L8: Entry 34 of 36 File: USPT Jul 17, 1990

DOCUMENT-IDENTIFIER: US 4942130 A
TITLE: T7 DNA polymerase

DEPR:
Modified T7 DNA polymerase efficiently utilizes dITP or deaza GTP in place of dGTP. dITP is substituted for dGTP in both the pulse and chase mixes at a concentration two to five times that a which dGTP is used. In the ddG chase mix ddGTP is still used (not ddITP).

Full | Title | Citation | Front | Review | Classification | Date | Reference | KWIC | Draw Desc | Image

---

☐ 35. Document ID: US 4921794 A

L8: Entry 35 of 36 File: USPT May 1, 1990

US-PAT-NO: 4921794
DOCUMENT-IDENTIFIER: US 4921794 A
TITLE: T7 DNA polymerase

AB: This invention relates to T7-type DNA polymerases and methods for amplification of DNA, for example by polymerase chain reaction.

L8: Entry 35 of 36 File: USPT May 1, 1990

DOCUMENT-IDENTIFIER: US 4921794 A
TITLE: T7 DNA polymerase

DEPR:
Modified T7 DNA polymerase efficiently utilizes dITP or deaza-GTP in place of dGTP. dITP is substituted for dGTP in both the pulse and chase mixes at a concentration two to five times that at which dGTP is used. In the ddG chase mix ddGTP is still used (not ddITP).

Full | Title | Citation | Front | Review | Classification | Date | Reference | KWIC | Draw Desc | Image

---

☐ 36. Document ID: US 4795699 A

L8: Entry 36 of 36 File: USPT Jan 3, 1989

US-PAT-NO: 4795699
DOCUMENT-IDENTIFIER: US 4795699 A
TITLE: T7 DNA polymerase

AB: This invention relates to T7-type DNA polymerases and method for using them.

L8: Entry 36 of 36 File: USPT Jan 3, 1989

DOCUMENT-IDENTIFIER: US 4795699 A
TITLE: T7 DNA polymerase

DEPR:
Modified T7 DNA polymerase efficiently utilizes dITP or deaza-GTP in place of dGTP. dITP is substituted for dGTP in both the pulse and chase mixes at a concentration two to five times that at which dGTP is used. In the ddG chase mix ddGTP is still used (not ddITP).

Full | Title | Citation | Front | Review | Classification | Date | Reference | KWIC | Draw Desc | Image

---

Generate Collection

Terms	Documents
l4 not l7	36

Display 50 Documents, starting with Document: 36

Display Format: TI,AB, Change Format

FILE 'CAPLUS' ENTERED AT [redacted]:48:42 ON 28 NOV 2001
L1 588767 S SEQUENC?
L2 889999 S DNA# OR RNA# OR NUCLEIC OR NUCLEOTIDE# OR POLYNUCLEOTIDE# OR
L3 316843 S L1 AND L2
L4 9 S DDITP OR (DIDEOXYINOSINE ADJ TRIPHOSPHATE)
L5 11 S DDITP OR (DIDEOXYINOSINE (W) TRIPHOSPHATE)
L6 1 S L3 AND L5

=> d l6 1 bib ab

L6 ANSWER 1 OF 1 CAPLUS COPYRIGHT 2001 ACS
AN 1994:451264 CAPLUS
DN 121:51264
TI The effects of nucleoside analogs on telomerase and telomeres in Tetrahymena
AU Strahl, Catherine; Blackburn, Elizabeth H.
CS Dep. Microbiol. Immunol., Univ. California, San Francisco, CA, 94143-0414, USA
SO Nucleic Acids Res. (1994), 22(6), 893-900
CODEN: NARHAD; ISSN: 0305-1048
DT Journal
LA English
AB The ribonucleoprotein enzyme telomerase is a specialized type of cellular reverse transcriptase which synthesizes one strand of telomeric **DNA**, using as the template a **sequence** in the **RNA** moiety of telomerase. The authors analyzed the effects of various nucleoside analogs, known to be chain-terminating inhibitors of retroviral reverse transcriptases, on Tetrahymena thermophila telomerase activity in vitro. The authors also analyzed the effects of such analogs on telomere length and maintenance in vivo, and on vegetative growth and mating of Tetrahymena cells. Arabinofuranyl-guanosine triphosphate (Ara-GTP) and ddGTP both efficiently inhibited telomerase activity in vitro, while azidothymidine triphosphate (AZT-TP), **dideoxyinosine triphosphate** (**ddITP**) or ddTTP were less efficient inhibitors. All of these nucleoside triphosphate analogs, however, produced analog-specific alterations of the normal banding patterns seen upon gel electrophoresis of the synthesis products of telomerase, suggesting that their chain terminating and/or competitive actions differ at different positions along the **RNA** template. The analogs AZT, 3'-deoxy-2',3'-didehydrothymidine (d4T) and Ara-G in nucleoside form caused consistent and rapid telomere shortening in vegetatively growing Tetrahymena. In contrast, ddG or ddI had no effect on telomere length or cell growth rates. AZT caused growth rates and viability to decrease in a fraction of cells, while Ara-G had no such effects even after several weeks in culture. Neither AZT, Ara-G, acycloguanosine (Acyclo-G), ddG nor ddI had any detectable effect on cell mating, as assayed by quantitation of the efficiency of formation of progeny from mated cells. However, AZT decreased the efficiency of programmed de novo telomere addn. during macronuclear development in mating cells.

=>